REMARKS

Following entry of this amendment, claims 1-4, 7, 9-11, and 13-17 will be pending in this application. Claims 5, 6, and 8 are canceled herein without prejudice, and claims 1, 2, and 7 are currently amended. Support for the amendments can be found throughout the application as filed, e.g., at original claims 5, 6, 8, and 12. No new matter has been added.

Restriction Requirement

Claims 2 and 7 were previously withdrawn by the Examiner as drawn to nonelected inventions. The Office action mailed August 18, 2009, asserted that claim 1 does not contribute a special technical feature over the alleged prior art publication Kanato et al., 2005, Biochem. Biopys. Res. Commun., 326:836-843 ("Kanato"). However, as noted in previously by Applicants, Kanato is not prior art against the pending claims. Applicants submitted with the previous reply a translation of the Japanese language priority document for this application, Japanese Patent Application Serial No. 2004-096744, which antedates the 2005 publication by Kanato. Perfection of the priority claim was acknowledged in the instant action. No additional examination burden would be involved in considering claims 2 and 7 as amended herein. Applicants therefore request withdrawal of the restriction requirement and examination of claims 2 and 7 with the currently pending claims..

35 USC § 103

Claims 1, 3-5, 9-17 were rejected as allegedly being unpatentable over EP 0846949 in view of Fraizer et al., 1995, Blood, 86:4704-06 ("Fraizer") and WO 97/39354 or Menssen et al., 1997, Blood, 89:3486-93 ("Menssen") or Baird et al., 1997, Exp. Hematol., 13:1311-12 ("Baird") or Loeb et al., 2003, Leukemia, 17:965-971 ("Loeb") or Tsuboi et al., 1999, Leukemia Res., 23:499-505 ("Tsuboi"). Claim 5 is canceled in this reply. Applicants respectfully traverse the rejection with regard to the remaining claims as amended herein.

The claims are amended herein to recite quantifying an expression level of the WT1 gene or reporter gene in a cell in a cell population within certain ranges of expression relative to

expression of the WT1 gene or reporter gene in a K562 leukemia cell and separating the cell based on that expression.[1]

EP 0846949 does not disclose such subject matter. Rather, EP 0846949 describes methods that use expression of WT1 in a population of $CD34^-$ cells as a marker of leukemic cells or solid cancer cells. EP 0846949 provide no disclosure relating to use of WT1 expression as a marker for hepatic, endothelial, or hematopoietic progenitor cells and no disclosure of the specific expression ranges recited in the claims.

Further, none of Fraizer, WO 97/39354, Menssen, Baird, Loeb, or Tsuboi cure the deficiencies of EP 0846949, as none of the publications disclose or would have made obvious the recited expression ranges.

Fraizer discloses a level of expression of WT1 in hematopoietic cells that is higher than that recited in the claims (see Table 1). Fraizer does not teach or suggest detecting the expression of a WT1 gene in a cell in a cell population in either of the recited expression ranges. Further, Fraizer does not teach or suggest the use of a reporter gene or detection of WT1 expression in a viable cell.

EP 0846949 is the national stage of the international application published as WO 97/39354 (see the cover page of EP 0846949, field (87)). WO 97/39354 therefore provides no additional disclosure that would remedy the deficiencies of EP 0846949.

Menssen discloses seeding of blood mononuclear cells onto agar plates and detection of WT1 gene expression in the resulting colonies by RT-PCR (p. 3486, left column). Menssen does not teach or suggest detecting the expression of a WT1 gene in a cell in a cell population in either of the recited expression ranges. Further, Menssen does not teach or suggest the use of a reporter gene or detection of WT1 expression in a viable cell.

Baird discloses isolation of $CD34^+$, $CD34^-$, and BMMNC cells using immunomagnetic beads followed by FACS (p. 314, right column) and sorting of cells into $CD34^+CD38^{-/lo}$ and $CD34^+CD38^{+/bright}$ populations by FACS (p. 315, right column). WT1 expression was then measured in the resulting populations using RT-PCR or immunohistochemistry in fixed cells. Baird does not teach or suggest detecting the expression of a WT1 gene in a cell in a cell

[1] A range of expression of $2.21 (\pm 1.62) \times 10^{-2}$ relative to expression in a K562 cell is indicative of a hepatic progenitor cell or an endothelial progenitor cell, whereas a range of expression of $3.54 (\pm 3.39) \times 10^{-4}$ is indicative of a hematopoietic progenitor cell. See the specification at page 8, lines 10-17.

population in either of the recited expression ranges. Further, Baird does not teach or suggest the use of a reporter gene or detection of WT1 expression in a viable cell.

Loeb discloses transfection of 32D cl3 cells with a plasmid expressing a WT1 isoform lacking both exon 5 and the KTS insert (designated WT1 (-/-)) under the zinc-inducible metallothionein (MT) promoter (p. 965, right column). WT1 expression was measured in the cells by western blotting, RT-PCR, and northern blotting (p. 965-966). Flow cytometry was used to detect expression of Gr-1 (a marker of mature cells) (Fig. 4) and for cell cycle analysis using Hoechst 33258 staining (Table 2). Loeb does not teach or suggest detecting the expression of a WT1 gene in a cell in a cell population in either of the recited expression ranges. Further, Loeb does not teach or suggest the use of a reporter gene or detection of WT1 expression in a viable cell.

Tsuboi discloses detection of differentiation markers Gr-1 and Mac-1 in cells constitutively expressing the WT1 gene (p. 502, left column). Tsuboi does not teach or suggest detecting the expression of a WT1 gene in a cell in a cell population in either of the recited expression ranges. Further, Tsuboi does not teach or suggest the use of a reporter gene or detection of WT1 expression in a viable cell.

Thus, without conceding that it would have been obvious to one skilled in the art to modify EP 0846949 based on Fraizer, WO 97/39354, Menssen, Baird, Loeb, or Tsuboi in the manner indicated by the Examiner, Applicants note that even if EP 0846949 were so modified, the result would not be the subject matter covered by claims 1, 3, 4, 9-12, and 13-17, as amended.

For at least these reasons, Applicants request reconsideration and withdrawal of the rejection of claims 1, 3, 4, 9-12, and 13-17.

CONCLUSION

Applicants submit that the pending claims are allowable and request early and favorable action thereon.

The fees in the total amount of $3160 for the RCE fee required under 37 CFR 1.17(e) ($810) and the Petition for Extension of Time fee ($2350) are being paid on the electronic filing system by way of deposit account authorization. Please apply any other charges or credits to Deposit Account No. 06-1050, referencing Attorney Docket No. 14875-0170US1.

Respectfully submitted,

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